AF
IAW

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No.:	09/829,113	Confirmation No.: 2302
Applicant(s):	William Edward Evans *et al.*	
Filed:	April 9, 2001	
Art Unit:	1637	
Examiner:	Jeffrey N. Fredman	
Title:	HAPLOTYPING METHOD FOR MULTIPLE DISTAL NUCLEOTIDE POLYMORPHISMS	

Docket No.: 044158/209598
Customer No.: 29312

Mail Stop Appeal Brief-Patents
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

REPLY BRIEF UNDER 37 CFR § 1.193(b)(1)

This Reply Brief is filed pursuant to 37 CFR §1.193(b)(1) and is filed in response to the Examiner's Answer of October 15, 2004, the Examiner's Answer being in response to an Appeal Brief filed August 24, 2004. Appellants note that the Answer acknowledges that claims 1-18, 21 and 22 do not stand or fall together.

APPELLANTS' CLAIMED INVENTION IS PATENTABLE OVER THE CITED REFERENCES

Generally, Appellants disclose a method for determining haplotype structure relative to distal polymorphisms using conventional techniques combined in a unique way . The method comprises the following steps set forth in claim 1:

(1) Obtaining a DNA sample comprising a contiguous DNA segment that encompasses distal polymorphisms.

(2) Using the DNA sample as a template for polymerase chain reaction (PCR) amplification of a DNA fragment comprising the DNA segment.

(3) Ligating the ends of said DNA fragment to each other so as to produce a circular DNA molecule, wherein the first nucleotide polymorphism and the second nucleotide

polymorphism are brought into closer proximity on the circular DNA molecule relative to the contiguous DNA segment.

(4) Determining the haplotype of the first nucleotide polymorphism and the second nucleotide polymorphism.

The key to this method lies in the second and third steps which provide a means for bringing distal polymorphisms on the same DNA strand in closer proximity to each other so that standard methods can be applied in the fourth step to determine their identity (i.e. haplotype).

Claims 1-11 and 13-16 have been rejected by the Examiner under 35 U.S.C. § 103 over Li *et al.* (1998) *BioTechniques* 25:358-361 in view of Patel *et al.* (1991) *Nucleic Acids Res.*, 19:3561-3567 and Michalatos-Beloin *et al.* (1996) *Nucleic Acids Res.* 24:4841-4843.

The Examiner incorrectly asserts that Li *et al.* teaches Appellants' second and third steps; i.e., producing a PCR fragment containing more than one polymorphism, then ligating the fragment to circularize the product and bring the polymorphisms into closer proximity on the circularized product. While Li *et al.* does describe the circularization of a PCR fragment containing more than one polymorphism, the process does not bring the polymorphisms into closer proximity.

The Examiner also incorrectly asserts that the secondary reference of Michalatos-Beloin *et al.* teaches the initial PCR step of the invention. Michalatos-Beloin *et al.* describes an allele specific long range PCR technique which is fundamentally different than the initial PCR step of the invention , which is not allele specific. In addition to failing to acknowledge this distinction, the Examiner also fails to acknowledge that Michalatos-Beloin *et al.* teaches away from its combination with other techniques. The advantage of the method described by Michalatos-Beloin *et al.* lies in the fact that it is a *single* step procedure in which the PCR reaction itself is used to determine haplotype. Further, even if combined with Li *et al.*, the Examiner fails to show how the combination would produce polymorphisms in closer proximity, as recited in Appellants' second and third steps.

The Examiner also cites Patel *et al.* for its teaching of inverse PCR from genomic DNA. This teaching is inapplicable because Appellants' claimed method does not depend upon inverse PCR (or any particular method of determining haplotype).

Because of these deficiencies, the rejection is improper and the subject matter of claims 1-11 and 13-16 is patentable over the cited references.

Besides the deficiencies enumerated above, the rejection of claim 12 is further defective: Claim 12 is also drawn to the method of claim 1 wherein the haplotype of the first nucleotide polymorphism and the second nucleotide polymorphism on the circular DNA molecule is detected by restriction fragment analysis of the circularized segment or of a PCR amplification product using the circular DNA molecule as a template. The Examiner has failed to cite any reference that shows utilizing such a technique after ligation and circularization. Rather, it is Appellants' own disclosure that provides this teaching. Because of this additional defect, the rejection of claim 12 should be withdrawn.

Claim 17 is rejected under 35 U.S.C. § 103 over Li *et al.* in combination with Patel *et al.* and Michalatos-Beloin *et al.* in further view of Krynetski *et al.* (1995) *Proc. Natl. Acad. Sci.*, 92:949-953. As described above, there is no motivation to combine Li *et al.* with Patel *et al.* and Michalatos-Beloin *et al.* Krynetski *et al.* merely teaches a point mutation of the TPMT gene and does not satisfy the deficiencies of the primary references. Because of these deficiencies, the subject matter of claim 17 is patentable over the cited references.

Claim 18 is rejected under 35 U.S.C. § 103 over Li *et al.* in combination with Patel *et al.* and Michalatos-Beloin *et al.* in further view of Martin *et al.* (2000) *Am. J. Hum. Genet.*, 67:383-394. There is no motivation to combine Li *et al.* with Patel *et al.* and Michalatos-Beloin *et al.* Martin *et al.* teaches SNPs in the region surrounding the APOE gene but does not satisfy the deficiencies of the primary references. Because of these deficiencies, the subject matter of claim 18 is patentable over the cited references.

Claim 22 specifies that DNA sequence immediately 5' to the first nucleotide polymorphism that encompasses an annealing site for a primer and DNA sequence immediately 3' to the second nucleotide polymorphism that encompasses an annealing site for a primer and that the length of these sequences is selected from less than 500, less than 400, less than 300, less

than 200, less than 100, or less than 50 bases. None of the references teach or suggest such a selection step. Because of these deficiencies, the subject matter of claim 22 is patentable over the cited references.

RESPONSE TO ARGUMENT RELATED TO CLAIMS 1-11 AND 13-16

The rejection of claims 1-11 and 13-16 under 35 U.S.C. § 103 is improper and should be withdrawn for the following reasons. First, the rejection mischaracterizes Appellants' claimed invention. Second, the rejection fails to apply the proper Section 103 standard. Third, the rejection is founded upon roughly five incorrect interpretations of the references—these incorrect interpretations appear throughout the Answer. Fourth, the Rule 132 declaration filed by Appellants has not been accorded its proper evidentiary weight.

I. The Rejection is Based Upon an Incorrect Interpretation of Appellants' Claimed Invention.

Appellants' claimed method includes a final general step for determining haplotype structure once the polymorphisms of interest have been brought into closer proximity to each other. While this step may utilize allele specific PCR amplication as a means of determining haplotype, it does not require such a process as there are many ways for determining haplotype once the polymorphisms are close to each other

Appellants have emphasized that this final step of the claimed method can be carried out without allele-specific amplification and that the independent claims do not recite any step using allele-specific amplification. See claim 1 and Appellants' Brief, page 3, ¶ 1, citing the specification, page 4, lines 8-12. Despite these clear statements, the Answer states that "[t]his invention is a form of ASIP (allele specific, inverse PCR), in which the only difference from the standard ASIP method is the distance between the first and second nucleotide polymorphisms." See the Answer, page 10, ¶ 2. The Answer compounds this misrepresentation of Appellants' invention again on page 14, lines 4-5 by stating that the "invention is the application of ASIP to polymorphisms spaced more than 200 base pairs apart...." To the contrary, there is no requirement that allele-specific PCR or inverse PCR be carried out to practice Appellants' claimed invention.

III. The Rejection is Based Upon Incorrect Interpretations Regarding the Teachings of the Cited References.

1. The steps of the method of Li et al. are NOT identical to the steps of Appellants' claimed method.

The Answer mistakenly asserts that "Li teaches a method in which the steps are precisely identical to those claimed." See the Answer, page 12, lines 9-12. Further, the Answer contends that "This fact is not disputed. The only difference resides in the preamble of the claim, where claim 1 has a requirement that the nucleotide polymorphisms of interest be separated by at least 200 nucleotides." *Id.* To the contrary, Appellants have repeatedly disputed the Examiner's mistaken assertion by demonstrating that the method of Li *et al.* is not identical to Appellants' claimed method. In particular, Appellants have shown that:

- Li *et al.* teaches a method that does not bring the relevant polymorphisms into closer proximity, as required by the third step of Appellants' claimed method. See Li *et al.*, Figure 1. Appellants have repeatedly explained this throughout prosecution. See Appellants' Brief, page 9, ¶ 3 to page 11, ¶ 2 and page 14, ¶ 2 to page 16, ¶ 2.
- Li *et al.* requires the application of allele-specific inverse PCR. There is no requirement that Appellants' claimed method be carried out by allele-specific amplification. See claim 1 and Appellants' Brief, page 3, ¶ 1, citing the specification, page 4, lines 8-12.

Despite Appellants' reiteration of the substantial differences between Li *et al.* and Appellants' claimed invention, the Answer maintains that the Li *et al.* method brings the polymorphisms into closer proximity. See the Answer, page 5, lines 6-7 and page 15, line 12. This is not so. The method of Li *et al. does not* generate a PCR fragment from a DNA segment to produce a circular DNA molecule wherein the polymorphisms are brought into closer proximity on the circular DNA molecule relative to the DNA segment, as required by all of Appellants' claims.

2. *The Examiner fails to explain how Li et al. would be modified to produce Appellants' claimed invention.*

The Answer also asserts that the final sentence of Li *et al.* would motivate the art worker to modify Li's method in an unspecified manner to produce Appellants' claimed method. This assertion appears repeatedly throughout the Answer on page 5, lines 6-7, page 12, lines 15-22, and page 15, lines 7-13. However, the Examiner fails to explain how Li *et al.* teaches or suggests modifying itself such that the polymorphisms of interest are brought into closer proximity on the circular molecule than they were on the DNA segment. As Appellants explained throughout prosecution, the circularization step of Li *et al.* fails to bring the polymorphisms into closer proximity. See Appellants' Brief, page 9, ¶ 3 to page 11, ¶ 3 and page 14, ¶ 2 to page 21, ¶ 2.

3. *Patel et al. does NOT provide motivation to modify the technique of Li et al..*

The Examiner asserts that Patel *et al.* suggest the application of Li's method to "haplotype polymorphisms up to 10,000 bases or more apart...." See the Answer, page 6, lines 8-10. To the contrary, the teachings of Patel *et al.* are not relevant to Li's method.

The method of Patel *et al. requires* restriction digestion to produce circularized genomic DNA, followed by relinearization of the genomic DNA, followed by inverse PCR. Li *et al.*, on the other hand, involves use of PCR to generate a template for inverse PCR and would not benefit from a step involving restriction digestion. Given the differences between the two methods, one of skill in the art would not be motivated to make the combination asserted by the Examiner. Further, although Li *et al.* and Patel *et al.* both use a form of allele-specific inverse PCR, Appellants' claimed method does not require allele specific amplification or inverse PCR. Thus, Patel *et al.*'s teachings regarding inverse PCR are simply irrelevant to the present invention. Appellants have reiterated these points throughout prosecution. See Appellants' Brief, page 12, ¶ 1 and page 21, ¶ 2 to page 22, ¶ 2. Nonetheless, the Examiner continues to rely on the teachings of Patel *et al.* related to inverse PCR. See the Answer, page 7, lines 20-21 and

page 11, lines 3-5. Such teachings are simply irrelevant to Appellants' claimed method, which does not require inverse PCR.

4. There is no reason to combine the one-step PCR technique of Michalatos-Beloin et al. with Li et al.

The Examiner has maintained that one would use the long-range PCR technique used by Michalatos-Beloin *et al.* to prepare template DNA for a haplotyping method, specifically the method of Li *et al.* See the Answer, page 7, lines 1-2 and 9-12. However, Michalatos-Beloin *et al.* teaches a one-step PCR technique where haplotyping is accomplished in the same PCR step by which hemizygous target DNA is isolated. See Appellants' Brief, paragraph spanning pages 12-13 and paragraph spanning pages 22-23. Thus, there is no reason to combine Michalatos-Beloin *et al.* with any other method, including the method of Li *et al.* The Examiner's conclusion also ignores the fact that the method of Michalatos-Beloin *et al.* produces a hemizygous product. As one of skill in the art would understand, a hemizygous product would be unsuitable for the method of Li *et al.*, which *requires* that a heterozygous product be produced in its first PCR step.

IV. The Errors Enumerated Have Resulted in an Incorrectly Formulated Obviousness Inquiry.

1. The accumulated errors have caused the Examiner to improperly formulate the issue on appeal.

As described above, the Examiner has mistakenly asserted that the method of Li *et al.* contains steps identical to the steps of Appellants' claimed invention. Based on this assertion, the Examiner asserts that the issue on appeal is whether it is "obvious to apply the ASIP method of Li to DNA templates where the polymorphisms are spaced more than the exemplified 30 nucleotide separation of Li?" In so doing, the Examiner ignores Appellants' second and third steps; i.e., producing a PCR fragment containing more than one polymorphism, then ligating the fragment to circularize the product and bring the polymorphisms into closer proximity on the circularized product. Although Li *et al.* circularizes a PCR fragment containing

more than one polymorphism, the polymorphisms were roughly 30-43 base pairs before circularization and are no closer than this after circularization. Consequently, Li *et al.* fails to teach or suggest a method involving the generation of a PCR fragment from a DNA segment to produce a circular DNA molecule wherein the polymorphisms are brought into closer proximity on the circular DNA molecule relative to the DNA segment.

2. *The Examiner has engaged in impermissible hindsight in formulating the rejection.*

Appellants have emphasized throughout prosecution that controlling caselaw requires the examiner to show by objective evidence of record a motivation to combine the references that create the case of obviousness to prevent the use of hindsight based on the invention to defeat patentability of the invention. See Appellants' Brief, page 6. The Examiner responds by stating that "[i]f the term 'hindsight' means anything in patent law, it must mean that the prior art references did not specifically desire the change which is the crux of the invention." See the Answer, page 14. To the contrary, the essence of hindsight is the combining prior art references without objective evidence of a suggestion, teaching, or motivation to combine the relevant teachings of the references. *In re Dembicazak*, 175 F.3d 994, 999, 50 U.S.P.Q.2d 1614, 1617 (Fed. Cir. 1999). Such an approach takes the inventor's disclosure as a blueprint for piecing together the prior art to defeat patentability. *Id.* The Examiner has failed to follow the rules against using hindsight to piece together the prior art. In addition, the Examiner relies upon subjective and incorrect statements regarding Appellants' invention and the cited references in support of the rejection.

The reliance upon subjective and incorrect statements is particularly evident on page 14 of the Answer. First, the Examiner states that "[t]he invention is the application of ASIP to polymorphisms spaced more than 200 base pairs apart." To the contrary, Appellants' claimed invention does not require allele specific amplification.

Second, the Examiner asserts that "Li states that ASIP 'can be applied to haplotyping of polymorphisms separated by a distance that is too long to be amplified by PCR..." and that this constitutes a teaching that suggests "the desirability of extending the distance of the ASIP

method." As stated above, Appellants' claimed method does not require ASIP. Further, Appellants' have demonstrated that if the quote from Li *et al.* is placed in context, it refers to a primer design problem caused by the 30-43 base pair separation between polymorphisms. Li et al., page 358, paragraphs spanning cols. 1-2 and page 361, col. 1. In any case, even if Li *et al.* were applied to polymorphisms separated by more than 30-43 base pairs, the Examiner fails to demonstrate how this would necessarily result in bringing the nucleotide polymorphisms into closer proximity on the circular DNA molecule relative to the contiguous DNA segment.

Third, the Examiner finds motivation for the combination of Li *et al.* with Patel *et al.* in Patel's statement that "having demonstrated DARMSI-PCR is feasible on relatively small regions, it is to be expected that the method should be applicable to regions as long as those amplified by conventional inverse PCR, i.e. 10 kb." See the Answer, page 14. This statement would not provide motivation for the combination of the dissimilar methods of Patel *et al.* and Li *et al.* As discussed above, the DARMSI-PCR technique of Patel *et al.* uses restriction digest of genomic DNA, followed by ligation, followed by relinearization, to prepare a template for subsequent PCR-based haplotyping by inverse PCR. The statement quoted by the Examiner indicates only that the PCR step of DARMSI-PCR should likely be capable of amplifying up to 10,000 base pairs and suggests only that the Patel *et al.* technique could be applied to polymorphisms separated by that distance. It fails to suggest the desirability of the combination of the restriction digest-based method of preparing a template for allele-specific PCR of Patel *et al.* with that of Li *et al.*, which involves the use of PCR to prepare gene-specific template for allele-specific inverse PCR. Furthermore, the Examiner has never explained how either technique should be modified to work with the other, nor has the Examiner explained how the asserted combination would produce Appellants' claimed invention.

Fourth, the Examiner finds motivation for the combination of references in the statement by Michalatos-Beloin *et al.* that "It should be possible to extend molecular haplotyping to distances up to 40 kb." However, this statement merely suggests that it should be possible to utilize the one step method disclosed by Michalatos-Beloin *et al.* for long distance haplotyping. This statement does not provide any suggestion or motivation to combine or modify this allele specific PCR method other than to utilize different DNA polymerases during PCR to minimize

template switching. Certainly there is no suggestion or motivation to add more steps to the method or to convert the allele specific PCR step to a non-allele specific PCR step as would be needed to arrive at the present invention.

In the present rejection, the Examiner has apparently used Appellants' claimed invention as a blueprint in an unsuccessful attempt to piece together disparate teachings from the prior art to arrive at the claimed invention with no motivation or suggestion to do so. . This is improper, and the rejection should be reversed accordingly.

V. The Examiner has failed to accord proper evidentiary weight to Appellants' Rule 132 Declaration regarding the teachings of Li et al.

In the final sentence of the publication, Li *et al.* summarizes their work by stating that "Although these alleles can be typed by allele-specific nested PCR following GPA-specific PCR, ASIP, rather than nested PCR, can be applied to haplotyping of polymorphisms separated by a distance that is too long to be amplified by PCR." During prosecution, the Examiner reasoned that "An ordinary practitioner would have been motivated to use long range PCR to prepare the template for the method of Li in order to extend the range of detection of polymorphisms in order to solve the problem of Li that there are 'polymorphisms separated by a distance that is too long to be amplified by PCR'." See the Office Action mailed 5/5/2004, page 6. The Examiner then asserted that "Michalatos-Beloin solves the problem using long range PCR." *Id.* Responsive to this statement, Appellants filed a Rule 132 Declaration explaining that the "distance problem" overcome by Li *et al.* was the roughly 30-43 bases that separated the nucleotide polymorphisms and prevented the design of a single allele-specific primer for use in the last step of the method (allele specific PCR-amplification). Read in context, the statement makes clear that the method of Li *et al.* was not a designed to solve the sort of distance problem amenable to long-range PCR and, further, does not produce a circular DNA molecule wherein the polymorphisms are brought into closer proximity on the circular DNA molecule relative to the DNA segment.

In the Answer, the Examiner now asserts that "When Li states that 'ASIP, rather than nested PCR, can be applied to haplotyping of polymorphisms separated by a distance that is too

long to be amplified by PCR...this statement cannot be taken as meaning anything other than bringing the polymorphisms close enough by the ligation of the ends so they can be analyzed by inverse PCR." However, Appellants' already provided evidence directly counter to the Examiner's conclusion in their Rule 132 Declaration:

> Li *et al.* concludes with the following statement: "Although these alleles can be typed by allele-specific nested PCR following GPA-specific PCR, ASIP [allele specific inverse PCR], rather then nested PCR, can be applied to haplotyping of polymorphisms separated by a distance that is too long to be amplified by PCR." Li *et al.*, page 361, column 1. When the statement of Li *et al.* is placed in context, it is clear that Li *et al.* refers to haplotyping polymorphisms that are too distantly separated for the design of a single forward PCR reaction. To place the comment in context, one must consider the following two points: (1) The only problematic distance discussed by Li *et al.* is a separation of 30-40 base pairs which causes a primer design problem for forward allele-specific PCR, necessitating the use of allele-specific nested PCR. Li *et al.*, page 358, column 2. (2) **Li *et al.* solves the problem by use of a circularization method that does not bring the polymorphisms closer** (see paragraph 8, above) followed by inverse PCR.

Declaration of Drs. McDonald and Evans, filed March 22, 2004, ¶ 9, quoted in Appellants' Brief, page 17 (emphasis added).

Rather than accord Appellants' declaration its proper evidentiary weight, the Examiner merely states that 'The Declaration reiterates the position of the inventors that in their scientific opinion the invention would not have been obvious...." This is not so. The Declaration contains specific evidence regarding the meaning and context of the Li *et al.* statement so heavily relied upon by the Examiner. This evidence is not the inventors' opinion regarding obviousness; it is the inventors' statement regarding the meaning of a scientific reference. It should be accorded proper weight as evidence that goes to the facts. The Examiner must rebut such evidence with his own evidence or concede the point. Despite the invitation to do so, the Examiner has not bolstered his rejection with evidence in the form of additional references or an affidavit under 37 CFR 1.104(d)(2) to support his conclusions, preferring instead his own subjective, out-of-context interpretation of the final sentence of Li *et al.* This is improper: a single line in a prior art reference should not be taken out of context and relied upon with the benefit of hindsight to

show obviousness. *See, e.g., Bausch & Lomb, Inc. v. Barnes-Hind/Hydrocurve, Inc.*, 796 F.2d 443, 230 U.S.P.Q. 416 (Fed. Cir. 1986).

For all of these reasons, the rejection of claims 1-18 and 21-22 should be withdrawn.

RESPONSE TO ARGUMENT RELATED TO CLAIMS 12, 17, 18, AND 22

The Examiner responded separately with respect to claims 12, 17, 18, and 22. Appellants note the following defects present in the response.

With respect to the rejection of claim 12, the Examiner asserts that Appellants argued that "there is no cited reference showing cleavage of the circularized segment by restriction analysis" and that Figure 7 of Patel *et al.* "expressly shows relinearization using a restriction enzyme..." Answer, page 17. Appellants actually stated "Claim 12 is also drawn to the method of claim 1 wherein the haplotype of the first nucleotide polymorphism and the second nucleotide polymorphism on the circular DNA molecule is detected by restriction fragment analysis of the circularized segment or of a PCR amplification product using the circular DNA molecule as a template. The Examiner has failed to cite any reference that shows utilizing such a technique after ligation and circularization." As explained above, Patel *et al.* teaches the following method: First, a restriction digest is carried out on genomic DNA isolated from an individual to be haplotyped. Second, the DNA fragments generated from the restriction digest are circularized using a DNA ligase. Third, the fragment is re-linearized utilizing a different restriction enzyme. Fourth, the haplotype is assessed by inverse PCR utilizing Amplification Refractory Mutation System primers. The relinearization step in Patel *et al.* is not used to *detect* the polymorphisms. Thus, the Examiner has failed to demonstrate that Figure 7 of Patel *et al.* cures the additional defect of the rejection of claim 12. The rejection should be reversed.

With respect to the defects present in the rejection of claims 17 and 18, the Examiner merely asserts that it would be desirable to haplotype the TPMT gene of claim 17 or the APOE gene of claim 18. See the Answer, page 18. However, as described above, there is no motivation to combine Li *et al.* with Patel *et al.* and Michalatos-Beloin *et al.* As applied to claim 17, Krynetski *et al.* merely teaches a point mutation of the TPMT gene and does not satisfy the deficiencies of the primary references. As applied to claim 18, Martin *et al.* teaches only the

presence of SNPs in the region surrounding the APOE gene but does not satisfy the deficiencies of the primary references. The defects remain uncured and the rejection of claims 17 and 18 should be reversed.

With respect to claim 22, Appellants' maintain that this claim (dependent upon claim 21) requires a selection step neither taught nor suggested by the cited references. Claims 21 and 22 are reproduced herein for the convenience of the reader (with the selection step in bold):

21. (Previously Presented) A method for determining the haplotype structure of a contiguous DNA segment comprising a first nucleotide polymorphism (NP) and a second NP separated by at least 200 nucleotides, said method comprising:

(a) obtaining a DNA sample comprising said contiguous DNA segment, wherein the DNA segment further comprises

a DNA sequence immediately 5' to the first NP that encompasses an annealing site for a primer and

a DNA sequence immediately 3' to the second NP that encompasses an annealing site for a primer;

(b) using said DNA sample as a template for polymerase chain reaction (PCR) amplification utilizing said primers of a DNA fragment comprising said contiguous DNA segment;

(c) ligating the ends of said DNA fragment to each other so as to produce a circular DNA molecule, wherein said first NP and said second NP are brought into closer proximity on said circular DNA molecule relative to said contiguous DNA segment; and

(d) determining the haplotype of said first NP and said second NP.

22. (Previously presented) The method of 21, wherein the DNA sequence immediately 5' to the first NP **has a length selected from** the group consisting of:

less than 500, less than 400, less than 300, less than 200, less than 100, or less than 50 bases long; and,

wherein the DNA sequence immediately 3' to the second NP **has a length selected from** the group consisting of:

less than 500, less than 400, less than 300, less than 200, less than 100, or less than 50 bases long.

Because claim 22 depends from claim 21, the selection step of claim 22 must produce a circularized PCR product wherein said first polymorphism and said second polymorphism are brought into closer proximity on said circular DNA molecule relative to said contiguous DNA segment. None of the cited references teach selecting an annealing site for the PCR primers sufficiently close to the polymorphisms of interest such that the circularization of the PCR

product will result in a product wherein the first polymorphism and said second polymorphism are brought into closer proximity on said circular DNA molecule relative to said contiguous DNA segment. The Examiner contends that Li *et al.* teach this step because it involves "a DNA sequence immediately adjacent to the 5' and 3' NPs which is less than 50 bases long." However, as discussed above, Li *et al.* fail to bring the polymorphisms closer together. Thus, the defect remains and the rejection should be reversed.

CONCLUSION

Appellants maintain that the Examiner has failed to carry his burden of establishing prima facie obviousness because he has (a) failed to establish the necessary suggestion or motivation in the prior art to combine the cited references in the manner proposed and (b) failed to articulate how the teachings of the cited references can be combined to arrive at the claimed invention. For these reasons, presented in detail in the Appeal Brief, Appellants respectfully requests that the rejections be reversed.

Respectfully submitted,

W. Murray Spruill
Reg. No. 32,943

CUSTOMER NO. 29312
ALSTON & BIRD LLP
Bank of America Plaza
101 South Tryon Street, Suite 4000
Charlotte, NC 28280-4000
Tel Raleigh Office (919) 862-2200
Fax Raleigh Office (919) 862-2260

CERTIFICATE OF MAILING

I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as first class mail in an envelope addressed to: Mail Stop Appeal Brief-Patent, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450, on December 13, 2004.

Polly P. Burton

Polly P. Burton